

240 County Road Ipswich, MA 01938-2723 Tel 978-927-5054 Fax 978-921-1350 www.neb.com info@neb.com

## New England Biolabs Product Specification

Product Name:	SHuffle® Express Competent E. coli
Catalog #:	C3028J
Shelf Life:	12 months
Storage Temp:	-80°C
Specification Version:	PS-C3028J v2.0
Effective Date:	01 May 2023

## Assay Name/Specification (minimum release criteria)

Antibiotic Resistance (Nitrofurantoin) - 15 µl of untransformed SHuffle® Express Competent *E. coli* streaked onto a LB or Rich Broth plate containing Nitrofurantoin will form colonies after incubation for 16 hours at 37°C.

Antibiotic Resistance (Spectinomycin) - 15  $\mu$ l of untransformed SHuffle® Express Competent *E. coli* streaked onto a LB or Rich Broth plate containing Spectinomycin will form colonies after incubation for 16 hours at 37°C.

Antibiotic Resistance (Streptomycin) - 15 µl of untransformed SHuffle® Express Competent *E. coli* streaked onto a LB or Rich Broth plate containing Streptomycin will form colonies after incubation for 16 hours at 37°C.

Antibiotic Sensitivity (Ampicillin) - 15 µl of untransformed SHuffle® Express Competent *E. coli* streaked onto a LB or Rich Broth plate containing Ampicillin will not form colonies after incubation for 16 hours at 37°C.

Antibiotic Sensitivity (Chloramphenicol) - 15  $\mu$ l of untransformed SHuffle® Express Competent *E. coli* streaked onto a LB or Rich Broth plate containing Chloramphenicol will not form colonies after incubation for 16 hours at 37°C.

Antibiotic Sensitivity (Kanamycin) - 15 µl of untransformed SHuffle® Express Competent *E. coli* streaked onto a LB or Rich Broth plate containing Kanamycin will not form colonies after incubation for 16 hours at 37°C.

Antibiotic Sensitivity (Tetracycline) - 15 µl of untransformed SHuffle® Express Competent *E. coli* streaked onto a LB or Rich Broth plate containing Tetracycline will not form colonies after incubation for 16 hours at 37°C.

**Phage Resistance** ( $\Phi$  80) - 15 µl of untransformed SHuffle® Express Competent *E. coli* streaked onto a Rich Broth plate does not support plaque formation by phage  $\Phi$  80 after incubation for 16 hours at 37°C.

**Functional Testing (Disulfide Bond Formation)** - The nuclease NucA requires disulfide bonds for its stability. When expressed at  $30^{\circ}$ C in *E. coli*, NucA is toxic to cells only in its oxidized disulfide-bonded state. Transformation of SHuffle® T7 Express Competent *E. coli* using 100 pg of plasmid that expresses a MBP-NucA fusion results in < 1% of the colonies when compared to a control transformation of its wild type parent strain NEB 10-beta after 0.4 mM IPTG induction at  $30^{\circ}$ C.



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**Transformation Efficiency** - 50  $\mu$ l of SHuffle® Express Competent *E. coli* cells were transformed with 100 pg of pUC19 DNA using the transformation protocol provided. Incubation overnight on LB-Ampicillin plates at 37°C resulted in >1 x 10e7 cfu/µg of DNA.

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Lauren Brown Quality Approver



Date 01 May 2023

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